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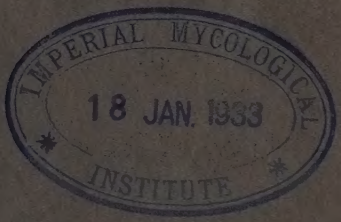
TWO DISEASES OF PEAS NEW TO ARIZONA

By
J. G. Brown and M. M. Evans



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TWO DISEASES OF PEAS NEW TO ARIZONA

By

J. G. Brown and M. M. Evans

INTRODUCTION

Diseases of green or garden peas and field peas are few in Arizona, but some of them can be very destructive. Two such diseases are new to the State. One of these is a foot rot caused by a species of Fusarium; the other is bacterial blight. The foot-rot disease is not described in any literature available to us, therefore we have studied it in considerable detail. To the excellent published description of bacterial blight (6) we add a few observations recorded in the work incidental to its identification in our material. Since the language used in this publication is necessarily more or less technical the diseases described, and other diseases of peas which occur in Arizona, will be generally discussed for the benefit of non-professional readers in a future bulletin of the Department.

PART I
FUSARIUM FOOT ROT

GENERAL

Foot and root rots of peas caused by Fusaria have been described from the established pea-growing districts of the United States and other countries, but no reference to these diseases in virgin soil has been found by the authors. The rot here described occurred in soil in which no cultivated crop had previously been grown. The field concerned was broken from virgin grassland in June, 1930, and planted to Dwarf Telephone and Stratagem varieties of peas about July 10 of the same year. By September 11 many plants had been killed, and at that time diseased specimens of the pea vines were sent to this laboratory.

ENVIRONMENT

The district in which the disease occurred, Chino Valley, is 17 miles north of Prescott. The elevation of the agricultural area directly concerned is approximately 4,400 feet. Although the precipitation is somewhat higher than that of the cultivated valleys of southern Arizona, irrigation is necessary. The soil is a highly alkaline, heavy loam with a moisture equivalent of 25 to 30 and an hydrogen-ion concentration averaging pH 8.6 to pH 8.8.*

* Figures supplied by Mr. E. S. Turville, County Agricultural Agent of Yavapai County and checked by the Department of Agricultural Chemistry, Arizona Agricultural Experiment Station.

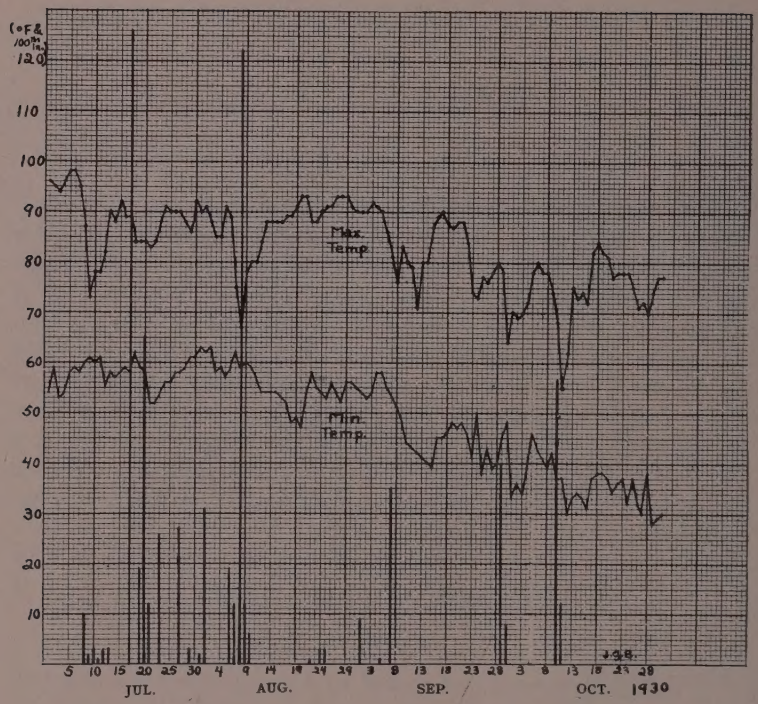

Fig. 1.—Temperatures in degrees Fahrenheit and precipitation in hundredths of an inch for the region in which Chino Valley is located and for the months in which foot rot of peas was active. Data on which the chart is based were reported by the United States Weather Bureau Station at Ash Fork, Arizona.

No meteorological data for Chino Valley are available, therefore those recorded at Ash Fork north of the Valley and about 700 feet higher are used. The temperatures at Ash Fork are probably somewhat lower and the precipitation somewhat higher than at Chino Valley. However, the figures given in the accompanying chart (Fig. 1) present a picture of temperatures and precipitation which prevailed in the general region of the disease at the time when the foot rot was active.

SYMPTOMS

Some entire plants received from the field were dead and brown. Others showed dead decayed roots and stem bases, and a varying number of yellow or brown leaves. Plants in the earlier stages of the rot had brown lesions of different sizes on stem and root in the region of the

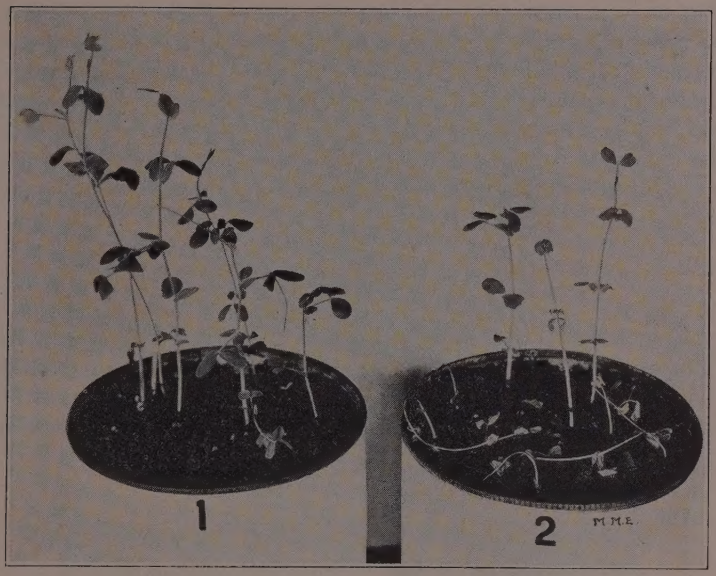

Fig. 2.—Canadian field peas inoculated with a monosporidial culture of Fusarium May 8, 1931; photographed May 27. 1. Control; 2. inoculated peas.

crown while the greater part of the stem retained its green color. In other plants the infection occurred chiefly on the root and extended scarcely above the crown. Little or no true wilting was evident in the field.

CAUSE

From the infected roots and stems of the peas a Fusarium was isolated which was studied in the following manner. Laboratory pots 5 inches in diameter were filled with soil and autoclaved for 2 hours at 15 pounds pressure. When cool, the soil in the pots was inoculated with a pure culture of the Fusarium and then planted with surface-sterilized pea seeds. Similar pots were kept uninoculated for control. The peas in the inoculated pots were attacked by the Fusarium, which caused marked dwarfing with lesions on taproot and stem base, resulting in death (Fig. 2). From the lesions the Fusarium was re-isolated.

Other pots prepared as described above were planted with surface-sterilized pea seeds which had been soaked after treatment in a suspension of Fusarium spores in sterile, distilled water. Still other pots

(Fig. 3) were inoculated with a mixed culture of *Rhizoctonia solani* and the Fusarium fungus which were originally isolated from the diseased specimens of pea vines received from the field. Peas planted in all inoculated pots were attacked, gave the symptoms shown by the peas in the field, and the Fusarium isolated from the field specimens and inoculated into the potted peas was easily obtained from the artificially inoculated peas. Mixed cultures affected the plantings more quickly than did pure cultures.

PURE CULTURES

The desirability of working with a monosporidial culture led to a trial of the capillary-tube method (1) for isolating single spores of Fusarium with which to start the culture. Spores from a potato-2 percent dextrose agar culture were used in a dilute suspension in the same agar. Capillary

Fig. 3.—Canadian field peas inoculated with a mixed culture of Fusarium and Rhizoctonia solani and planted September 29, 1930; photographed October 14. 1. Control; 2. inoculated peas.

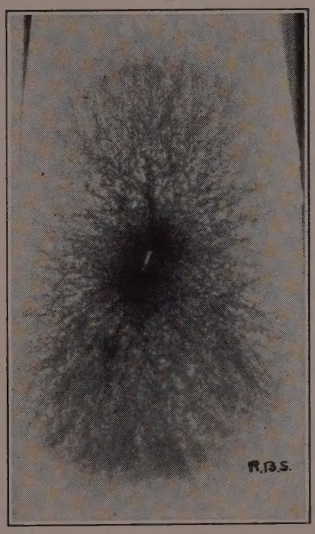

Fig. 4.—Young monosporidial culture of Fusarium from foot rot of peas, on potaro-2 percent dextrose agar. Note small piece of glass tube in center in which was isolated the single spore from which the mycelium grew. x 3.

tubes were drawn from 5-mm. glass tubing. After the capillary had been dipped in 95-percent alcohol as a precautionary measure against accidental contamination, it was fitted at the large end with a depressed rubber syringe bulb. The capillary was then immersed in the warm agar suspension of Fusarium spores, filled with agar through the action of the bulb (which was then removed), and placed on clean filter paper for a few moments in order to solidify the agar. With the high-power dry lens of the microscope the spores of the Fusarium in the agar of the capillary were now located and the capillary was cut into pieces which contained a single spore each. The individual pieces of the capillary were then dipped in a 1:1000 solution of mercuric chloride, rinsed in sterile, distilled water, and dropped singly on agar slants. Germination of the spore in each piece of capillary gave an unquestionable monosporidial culture (Fig. 4). This was the source from which seedings were made on the following media, and all the cultures whose physiological reactions are described were derived from one and the same spore.

REACTIONS ON MEDIA

Autoclaved Rice

Plates Seeded from Monosporidial Culture

28° C.	37° C.
24 hours	
Tuft of delicate, erect, pale, ochraceous-buff,* aerial hyphae; pionnotes on one plate, cinnamon-buff.	No growth.
4 days	
Pionnotes 9 mm. in diameter to 8 × 23 mm., moist, glistening, honey yellow; no aerial hyphae in two-thirds of cultures, in others a small (3 mm. diameter) tuft of gray, delicate, erect hyphae on inoculum only.	No growth.
Cultures flat, moist, with scant aerial hyphae; some granular, others velvety with fine, closely interwoven hyphae; 1 cm. to 4.5 cm. in diameter.	No growth.
15 days	
Pionnotes covers entire upper surface of substrate (93 mm. diam.), conforms to outlines of rice grains, raw sienna on margin to antique brown center; thin growth of aerial hyphae from center to 1 cm. of margin, light vinaceous-fawn surrounding grayer pulverulent center; under side of substrate has spots of light orange-yellow.	Pionnotal growth scant, 25 × 34 mm., moist, glistening, cream color; no aerial hyphae; no growth on under side of substrate.
21 days	
	No change.

Autoclaved Yellow Corn Meal

Plates

28° C.	37° C.	Room temperature
	24 hours	
		Growth of very fine, loose aerial hyphae; 4-8 mm. in diameter, gray to pale cinnamon-pink; no discoloration of substrate.
	48 hours	
		Growth scant, 7-13 mm. in diameter, pale pink-tinged gray.

* Ridgway, Robert.— Color Standards and Color Nomenclature, Washington, D. C., 1912.

4 days

Pionnotes 20 × 27 mm., pale grayish vinaceous except spot 5 × 10 mm., avellaneous to wood brown; no aerial hyphae; lower surface of substrate clay to tawny olive.

Pionnotes 3-5 × 12 mm., shell pink; no aerial hyphae; no growth on under surface of substrate.

7 days

Flat, thin pionnotes 33-36 × 37-44 mm.; margin a very narrow zone, wood brown to vinaceous-buff; next inward zone 3-4 mm., phlox pink; next zone 3 mm. wide, pale amaranth pink; central zone color of the corn meal; aerial hyphae scant

10 days

Pionnotes 46 × 55 mm.; outer part pale vinaceous-fawn and vinaceous-gray to walnut brown; central part buff-pink to walnut brown; under surface of substrate buckthorn brown to dresden brown; scant growth of arachnoid aerial hyphae in spot 12 × 20 mm.

No change.

Pionnotes 50-52 × 57-60 mm., granular like substrate; gradual transition in color, outside inward: clay, pale lilac, pale vinaceous-pink, light buff or shell pink in center; sometimes roundish spot or crescent of hydrangea red on side; aerial hyphae central, scant; lower surface of substrate isabella around cartridge buff, or isabella.

14 days

Margin light vinaceous-gray to light vinaceous-lilac; central part covered with fine felt of olive-buff to purplish-vinaceous aerial hyphae; lower surface of substrate has marginal mycelium of pale vinaceous-fawn.

No change.

Autoclaved White Corn Meal

Plates

28° C.

37° C.
24 hours

Room temperature

Growth of fine, loose aerial hyphae, gray to pale orange-yellow, 6-12 mm. in diameter; no discoloration of the substrate.

48 hours

Aerial hyphae scant, faintly tinged with pink, in area 7-16 mm. in diameter; pionnotes tawny-olive to clay.

4 days

Growth 12 × 28 mm., buckthorn brown to Prout's brown; no aerial hyphae, no growth on lower surface of substrate.

Growth 3 × 12 mm., pale vinaceous-fawn; no aerial hyphae; no growth on lower surface of substrate.

7 days

Flat, thin pionnotes 32-40 × 39-49 mm.; marginal zone 2 mm. wide, vinaceous-buff, clay to wood brown; next band phlox pink, 2-3 mm.; band, hydrangea red to magenta, 10-14 mm.; center chamois to cinnamon-buff. Tuft of aerial hyphae, center, pinkish buff to tawny-olive.

10 days

Pionnotes 49 × 52 mm., light pinkish cinnamon in spots, but mostly isabella to Saccardo's umber; lower surface of substrate delicately tinted pinkish.

No change.

Pionnotes 36-62 × 59-67 mm., granular like substrate; color transition outside inward: clay to vinaceous-buff, pale lilac, pale vinaceous-pink, hydrangea red to Hay's maroon (broadest), cinnamon-buff (center); aerial hyphae central, grayish to pale brownish vinaceous; growth scant on under side of substrate, isabella to tawny-olive.

Autoclaved Potato

Slices, 28° C.

Plugs, 27° C.

24 hours

Scant growth of short, fine aerial hyphae, light vinaceous-fawn.

48 hours

Pionnotes 6-20 mm., pale pinkish buff to pinkish buff; slight to fair growth of aerial hyphae, light pinkish cinnamon.

4 days

Pionnotes covers half of surface of slant, smooth, moist, glistening, salmon-buff; no aerial hyphae; no odor.

6 days

Pionnotes dull, moist, smoth, pinkish buff to light ochraceous-salmon, covers surface of substrate; no aerial hyphae; potato discolored smoke gray to hair brown.

Pionnotes light ochraceous-salmon.

7 days

Red color appears in 1 culture.

11 days

Pionnotes pale pinkish buff to light ochraceous-salmon to vinaceous-pink in center, eugenia red on margin and lower surface of substrate; surface dull and granular in some cultures, moist, glistening and rugose in others; aerial hyphae fine, sparse, pale pinkish buff, present in a few cultures.

18 days

(29° C. since seeding)

Pionnotes somewhat leathery, glistening, rugose, hydrangea red on ridges and vinaceous-pink in hollows; aerial hyphae none or more commonly a small delicate tuft, rarely completely covering pionnotes ,white, pale gull gray, pale vinaceous or pale pinkish buff.

PROTEOSE-PEPTONE AGAR*

Plates—27° C.

48 hours

pH 5.2

Growth 6-17 mm. in diameter, dull, moist; margin pale olive-buff, consists of very fine hyphae closely appressed to medium; zone of pale cinnamon-pink from margin to inoculum in center; inoculum covered with salmon-buff pionnotes. No difference noticeable between colonies.

pH 7.00

Same as for pH 5.2.

4 days

Growth 20-22 mm. in diameter; margin 3 mm. dull, pale cinnamon-pink, of fine hyphae as above; next region 3-4 mm. wide, granular, moist, glistening, honey yellow to yellow ocher, consists of filaments in process of immersion; center yellow ocher to buckthorn brown, somewhat raised and rugose pionnotes, moist, glistening.

Growth 20-22 mm. diameter, mostly dull; margin 3 mm., similar in color to opposite but hyphae coarser; central parts changing from yellow ocher to eugenia red, flat, very finely granular.

* Leonian, Leon H.— Studies on the variability and dissociations in the genus Fusarium. Phytopath. 19: 769, 1929.

7 days (Plate IV, Figs. 37, 38)

Growth 26-36 mm. in diameter, dull, finely granular, faintly radiate and circularly zonate near margin; margin 1-2 mm., pale cinnamon-pink; central part from sulphine yellow in narrow outer belt to cinnamon; no aerial hyphae.

Growth 30 mm. in diameter; margin 1-3 mm., dull, moist, pale, cinnamon-pink, more coarsely lobate than on pH 5.2; central part mostly acajou red, but lighter band 1½ mm. wide and 5 mm. from margin.

Potato—2 Percent Dextrose Agar

Slants—28° C.

4 days

pH 5.6

Most of surface thinly covered with dull, moist, tough pionnotes; needle-track a dark vinaceous, slightly elevated ridge 1 mm. wide from which radiates a delicate, dark vinaceous filamentous network which becomes gray in the almost invisible margin; small cinnamon-buff spot surrounds minute (1 mm.) piece of inoculum in needle-track.

pH 7.00

Growth somewhat greater than at pH 5.6; same color but proportion of gray hyphae greater.

14 days

Middle to base of ridge in needle-track becoming rugose; growth showing on lower surface of substrate. Upper surface hydrangea red to mineral red, with spot of light ochraceous-buff at base of slant in some tubes; growth on under surface of substrate ox-blood red; aerial hyphae in a few tubes.

Growth markedly greater than at pH 5.6, but color the same; short, fuzzy, light vinaceous-gray aerial hyphae on pionnotes in 1 tube only.

CZAPEK'S SOLUTION AGAR, MODIFIED*

Plates, pH 5.8

28° C.	37° C.
24 hours	
Gray pionnotes 2 mm. in diameter.	No growth.
6 days	
Growth scant as compared with other media; central pionnotal part 18 mm. in diameter, dull, moist, pinkish buff, or pinkish buff radially streaked with gray; marginal halo of very fine, gray mycelial filaments, 14 mm. wide.	No growth.

* Thom and Church.—The Aspergilli, p. 39. 1926.

MALT EXTRACT AGAR*

Plates

Reactions (not adjusted) pH 4.8; 27-28° C.

48 hours

Growth 13 mm. in diameter, flat convex to umbonate, radiately filamentous in outer part; margin entire or faintly crenulate, thin, pale pinkish cinnamon; pionnotes same color, 1 mm. in diameter covering and surrounding inoculum.

7 days

Growth 22-29 mm. in diameter; margin entire or rhizoid with a conspicuous gray halo; pionnotes flat to umbonate, smooth to distinctly contoured, pale cinnamon-pink to buff-pink.

AUTOCLAVED WALNUT STEM

27° C.

6 days

Growth mainly fine, delicate, branching hyphae creeping over surface of substrate and glass wall of tube and becoming immersed to form pionnotes, pale cinnamon-pink and pale pinkish buff to verona brown and liver brown; small tuft of erect aerial hyphae at point of inoculation; aerial hyphae appear gray but prove to be pale pinkish buff to pale cinnamon-pink.

AUTOCLAVED ROSE STEM

27° C.

6 days

Like above.

LIVING WHOLE POTATO TUBER

27° C.

21 days

Very slow growth (3 mm. diam.) of fine, cottony, aerial hyphae, pale pinkinsh buff, and minute pionnotes, pale cinnamon-pink. Potato discolored brown for 1-2 mm. around needle-puncture.

MYCELIUM

The mycelium was studied not only in the various media as previously described, but also in stained sections of infected peas and similarly prepared sections through cultures on autoclaved potato plugs. In preparing the latter, a slice was made parallel to the flat surface of the plug which, with its covering of mycelium, was afterwards dropped into chromo-acetic acid. After killing and fixing were complete the material was washed, dehydrated, cleared, embedded, sectioned, and stained in Haidenhain's iron-alum haematoxylon.

Stained sections of Canada field peas were made from young plants which had been grown from surface-sterilized seeds in steam-sterilized

* Leonian, Leon H.—Studies on the variability and dissociations in the genus Fusarium. Phytopath, 19: 769. 1929.

pots and soil, and which had been inoculated by adding to the soil spores from a monosporidial culture of Fusarium. When the pea vines were 6 to 8 inches high the bases of the plants were prepared for sectioning. The sections, cut lengthwise through the base of the stem and the adjacent part of the root, showed masses of the hyphae on the surface of the latter. Under the hyphae, the cells of the epidermis and cortex were either killed or injured. With carbol-fuchsin the dead cells stained a very deep red and the injured cells a lighter red, which distinguished them from cells which were healthy at the time that the blocks of pea plant were dropped into the killing fluid, since the healthy tissues retained less color. Dead cells forming the nucleus of the lesion contained parts of one or more hyphae, while the injured marginal cells had not been invaded but appeared to have been affected by some diffusing toxic substance.

The hyphae were both inter- and intra-cellular (Fig. 5) in the epidermis and cortex. The part of a hypha which passed through a cell wall was often, but not always, reduced in size with subsequent enlargement just inside the invaded cell (Fig. 5, c). From the enlargement one or more filaments which varied in size usually were branched off. Frequently the cells of the host contained several hyphae which traversed one or more host cells. No invasion of the stele was noted in the young plants, and the infection appeared to work from the outside inward.

The mycelial filaments are fine in the very young culture and in the margin of older cultures. They are simply branched (Plate I, Fig. 1). Immersion of the mycelium and the formation of pionnotes begin in the center of the culture and progress peripherally (Plate IV, Figs. 36, 37, 38, 41, 42). The usual anastomoses of hyphae (Plate I, Fig. 2) as well as spores (Plate I, Figs. 8, 9), and hyphae with spores (Plate I, Fig. 10) occur. Filaments become coarser. The interlacing and anastomoses of the hyphae and the adhesion of the conidia form the thin layer consisting of stromatal bed and pionnotes, respectively. The latter may or may not have some aerial hyphae on its surface (Plate IV, Figs. 38, 39, 40). Immediately underneath the layer of spores there is a thin, loose network of hyphae which rests upon and is continuous with a denser, thicker stromatal layer (Plate IV, Fig. 43). In the potato medium itself the filaments are relatively sparse.

Many of the hyphae under the pionnotes show pale in stained sections through the potato medium. Evidently they gradually lose their cell-contents and finally appear optically empty when an attempt is made to stain them. At the time there is still an abundance of food in the medium. Either food is withdrawn from the hyphae faster than it can

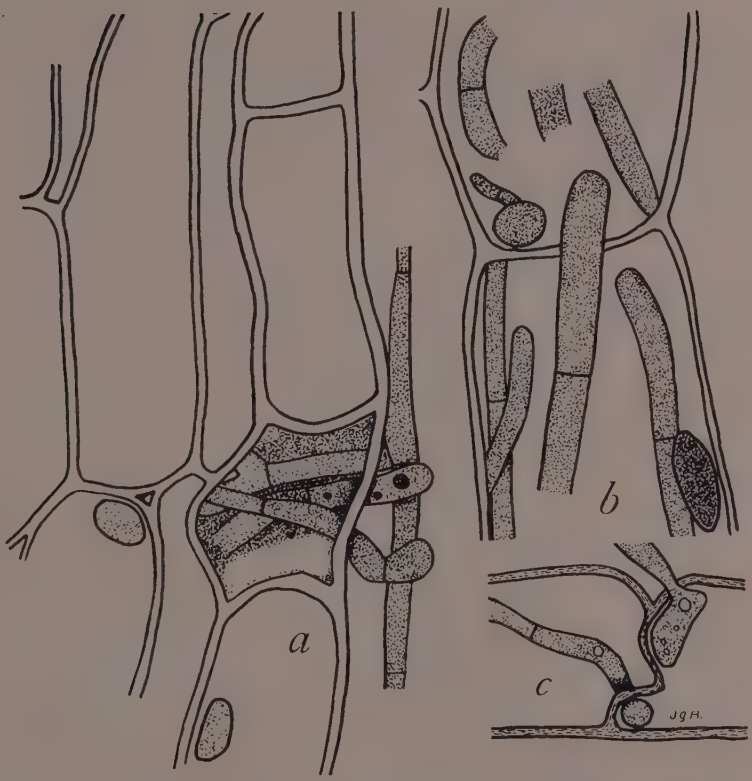

Fig. 5.—Relation of the hyphae to the host cells in Fusarium foot rot of peas. a. Epidermal cell of root attacked by the fungus. x 803. b. Parts of two cortical cells of the root containing hyphae; drawn without change of focus of microscope. x 1560. c. Parts of two cortical cells of the root showing enlargement of hypa within the cell. x 1560.

be replaced from the substrate, or else a toxic substance is formed which injures the hyphae. Enlarged cells which appear to be of two kinds with reference to their fate, are common in this region: one kind becomes extensively vacuolate, loses its nucleus, shows a network of cytoplasm which sometimes resembles a nuclear reticulum, and finally is optically empty like the smaller cells previously described (Plate III, Fig. 35, a, b, c); the other kind has abundant protoplasmic and food content (Plate I, Figs. 3; 4, a, b; 5) and develops into a resting cell which may or may not show the wall structure of a true chlamydospore.

Diameters of 20 vegetative cells in the stromatal region on potato plug, in stained sections, ranged from 2.08μ to 3.58μ and averaged 2.73μ. In the same region the normal vegetative cells had nuclei averaging 1.32μ $\times$ 1.15μ, the largest measured being 1.62μ $\times$ 1.85μ and the smallest 0.92μ $\times$ 1.15μ.

Neither sclerotia nor sporodochia were found either in cultures or stained sections.

SPORES (PLATES II, III)

Microconidia and macroconidia appear in cultures. The microconidia vary much in form and size. Although little difficulty was experienced in finding microconidia in the cultures, they are not abundant.

Macroconidia were dorsiventral, mostly apedicellate, varying in color with the pionnotes as previously described under cultures. Occasionally the color was sufficiently intense to show in a few individual spores under the highest power of the microscope, for instance in the case of spores from autoclaved potato plugs and autoclaved rice. When masses of the red spores or yellow spores were centrifuged after killing in chromo-acetic and washing with distilled water, some of the color diffused out into the alcohols used for dehydrating. Giant spores (Plate III, Fig. 30) were common in some cultures; also elongated hyaline spores without septa (Plate II, Fig. 15) or with indistinct septa.

Spores resembling chlamydospores (Plate I, Fig. 4, a) were common in all cultures but they showed only a single wall. However, true chlamydospores were present in old cultures (Plate III, Figs. 33, 34). Both types of cells were intercalary and terminal; the chlamydospores also occurred in clusters (Plate III, Fig. 34). Only a few chlamydospores were measured, which ranged in size from 7μ to 18μ.

REACTIONS TO INOCULATIONS

a. Seeds. Pea seeds of the variety, Premium Gem, soaked in a heavy suspension of spores of the Fusarium fungus, became severely infected. On the surface of the moist seeds the cotyledons were hydrangea red to mineral red; between the cotyledons the color was seashell pink to light ochraceous-buff. When planted in autoclaved soil the seeds gave only a 75-percent stand against the 90- to 95-percent stand obtained in the controls.

b. Young plants. Pea seedlings were grown from surface-sterilized seeds in autoclaved soil and inoculated through the soil, or by dipping the seeds in a suspension of Fusarium spores in sterile, distilled water.

MEASUREMENTS OF SPORES* FROM CULTURES DERIVED FROM A SINGLE SPORE.

Type	0—Sept.	1—Sept.	2—Sept.	Autoclaved Corn Meal—39-40 days 3—Sept.	4—Sept.	5—Sept.	6—Sept.
No. meas'r'd	10	10	10	10			
Size	7.81-13.79×3.22-5.51	10.57-20.69×3.22-4.14	17.93-25.52×3.68-4.60	29.43-45.52×3.22-4.60			
Average	10.39×4.07	16.57×3.66	21.20×4.21	38.44×3.82			
				Same medium—111 days			
No. meas'r'd	1	4	1	19	18	8	
Size	12.35×2.74	13.72-18.30×3.20-3.88	20.81×2.97	21.95-31.10×3.20-4.57	27.90-35.63×2.97-4.57	31.10-41.17×3.65-4.10	
Average		15.32×3.42		27.95×3.74	31.39×3.76	35.70×3.98	
				Same medium—324-326 days			
No. meas'r'd		5	5	11	35	35	1
Size		14.18-16.46×2.51-3.88	15.55-24.47×3.20-4.11	14.41-38.42×2.74-4.34	25.38-50.78×2.74-4.57	21.50-55.80×2.74-4.57	46.66×3.43
Average		14.90×3.94	19.11×3.56	26.13×3.32	36.13×3.44	41.31×3.79	
				Autoclaved Potato Plug—11 days			
No. meas'r'd	1	1	2	2	2	3	2
Size	12.72×3.63	15.90×3.63	23.40-25.90×2.72-3.18	15.90-21.14×3.18-3.63	35.00-40.50×3.63-4.10	37.30-40.45×3.63	41.10- 48.00×4.50-5.05
Average			24.65×2.94	18.52×3.40	37.75×3.86	39.09×3.63	44.55×4.77
				Potato-2% Dextrose Agar, pH 7.00—20 days			
No. meas'r'd	12	6	2	23	6	14	3
Size	6.79-19.47×2.49-3.21	11.03-31.25×2.52-5.20	17.01-19.31×3.44-3.67	22.98-46.64×2.76-4.75	31.72-45.05×3.21-4.13	37.70-56.55×3.67-4.59	59.31-104.00×4.13-11.0
Average	13.44×2.82	20.17×3.68	18.16×3.55	36.37×3.85	38.59×3.84	47.03×4.02	84.60×8.37
				Proteose-Peptone Agar, pH 7.00—13-14 days			
No. meas'r'd	11	20	24	31	28	5	
Size	7.60-17.97×2.30-3.68	10.82-25.11×2.53-4.37	14.74-37.32×2.99-5.06	21.42-44.00×2.99-4.83	32.25-53.91×2.30-4.60	43.77-49.30×3.68-5.53	
Average	12.43×2.89	18.79×3.42	24.35×3.53	33.46×3.75	41.26×3.76	46.07×4.14	
				Summary			
No. meas'r'd	35	46	44	96	89	65	6
Size	6.79-19.47×2.30-5.51	10.57-31.25×2.51-5.20	14.74-37.32×2.72-5.06	14.41-46.64×2.71-4.83	25.38-46.08×2.30-4.60	21.50-56.55×2.74-5.53	41.10-104.00×3.43-11.0
Average	12.20×3.36	17.71×3.49	23.11×3.65	32.39×3.76	37.15×3.68	42.82×3.88	64.93×8.18

* In microns; made with ×15 apochromatic ocular and 2-millimeter apochromatic objective, N.A. 1.40.

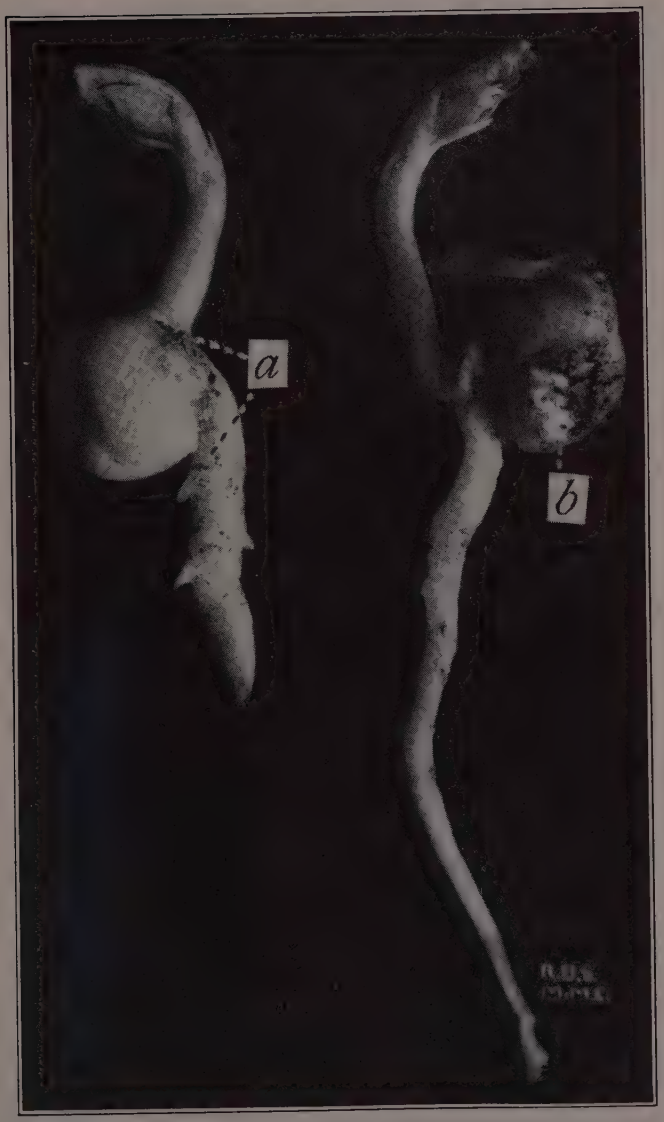

Fig. 6.—Fusarium foot rot of seedlings of Canadian field peas grown from surface-sterilized seed in sterilized, inoculated soil. The roots are in an early stage of the disease and the cotyledon, b, in a more advanced stage. a. Brown, freckle-like lesions; b. mycelium of Fusarium on cotyledonary lesion.

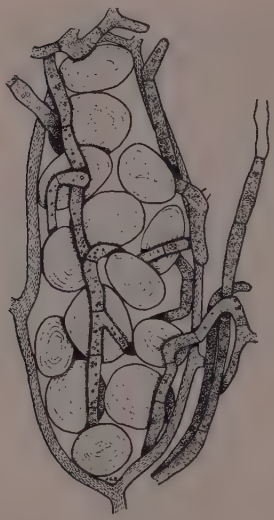

Fig. 7.—Cell from lesion like that shown at "b," Figure 6, in cotyledon of pea. Protoplasmic contents are gone but starch grains are intact. x 390.

Dwarfing of the resulting plants has already been mentioned. Lesions occurred on the cotyledons, taproot, and stem base. They were small, roundish, freckle-like brown spots (Fig. 6). The lesions were most numerous on the young taproot in the region of the cotyledons where a considerable growth of mycelium usually was evident (Fig. 6, a). On the cotyledons deep ulcers were produced (Fig. 6, b). Cells from the cotyledonary lesions when placed under the microscope, showed complete destruction of the protoplasmic contents (Fig. 7). The lesions on the taproot rapidly increased in size, coalesced, and extended upward on the stem (Figs. 8, a; 9, b) and downward on the root. The taproot and stem base were destroyed (Figs. 8; 9, b, c).

Superficially the disease on the pea seedlings resembled rhizoctoniose, but careful examination revealed differences. The lesions on the taproot in the foot-rot disease were rounder and smoother as compared with the more irregular lesions in rhizoctoniose. Cotyledonary lesions afforded a greater contrast. In foot rot they are deep as compared with the superficial lesions of rhizoctoniose (Fig. 10). The presence of sclerotia in seedlings attacked by Rhizoctonia (Fig. 10, b) appears to be a distinguishing character, since no sclerotia were found either in cultures of the Fusarium or in plants infected with the foot rot.

Fig. 8.—Fusarium foot rot on seedling of Canadian field pea. The taproot is in an intermediate stage of the disease. Inoculation of sterilized soil, October 30; photographed November 6. a. Lesion with mycelium on taproot and cotyledon. Other younger lesions are evident.

Fig. 9.—Advanced stages of Fusarium foot rot in seedlings of Canadian field peas. a. Lesions which have almost severed the taproot; b. lesions progressing upward on the stem.

c. Older plants. The leaflets and stipules of older plants turned yellow, curled downward, became lifeless and dry. The stems turned light green to light brown and finally died. Stem bases were decayed an inch or more above the soil line and the decayed region was brown in color.

IDENTIFICATION OF THE FUSARIUM

The presence of a true pionnotes, its colors, and the shape and nature of the spores place the Fusarium which is described in this paper in the

Fig. 10.—Comparison of Rhizoctonia root rot (1) and Fusarium foot rot (2) of pea seedlings. a. Mycelium of Rhizoctonia on discolored cotyledons; b. lesions caused by Rhizoctonia with two sclerotia on seedling at extreme left; c. Fusariose of cotyeldons; d. lesion caused by Fusarium on taproot.

Section Eupionnotes, Sub-section Chlamydospora. We believe that the fungus is a previously undescribed variety of *Fusarium merismoides* Corda (8).

CONTROL

The fact that the foot rot of peas occurred in virgin soil suggests that the disease is seed-borne. On the other hand no Fusarium of the type described appeared in cultures of seeds obtained from the seed house which supplied the peas planted in the field affected with the rot. This may mean nothing. None of the seed purchased for use in the Chino Valley was procurable for study; therefore we cannot be sure that the seed used was clean. If infection were present it was probably internal, for the seed used was treated with Semesan Bel before planting.

Although the appearance of pea plants inoculated in the laboratory and greenhouse indicates that infected plants would be killed before the fungus could reach the pods and infect the young seeds, some of the naturally infected plants from the field which were alive when received at the laboratory carried pods. Further investigation is necessary in order to determine the possibility of seed infection and dissemination.

In the meantime the general rules for the control of similar diseases should be observed. These include the selection and treatment of the best seed, the planting of fields not cropped to peas within a period of 4 or 5 years, rotation of crops, and field sanitation. The ideal solution of the problem is, of course, a rot-resistant variety of pea. Varieties resistant to Fusarium wilt have been reported (4), but their reaction to the Fusarium herein described has not yet been determined.

PART II

BACTERIAL BLIGHT

GENERAL

Bacterial blight of peas was first described from Colorado in 1915 by Dr. Walter G. Sackett (6). It had appeared the previous year in field peas in the San Luis Valley and also in garden peas in a distant and lower part of the State. It was worse on peas growing on sandy soil where the supply of moisture was limited. Crops on new ground and on previously cultivated ground were alike affected. The disease attacked White Mexican, Warshauer, and Alaska varieties. Prior to Sackett's work the disease had been attributed to various causes.

GEOGRAPHICAL DISTRIBUTION

Since Sackett's recognition of the disease, bacterial blight has appeared in several states. According to the United States Department of Agriculture (9) the disease has been found in Colorado, Delaware, Florida, Illinois, Indiana, Maryland, Michigan, Montana, New York, North Dakota, Oregon, Pennsylvania, South Carolina, and Wisconsin. The blight has also been reported from California.

LOSSES

Severe infection with bacterial blight may reduce the stand of young peas one-third, according to Sackett. Jones and Linford (3) state that early infections in spring, when the plants are yet young, may seriously reduce the stand, and that serious infection at any time later may weaken the plants, at times destroying the entire leaf surface. Jennison (2) estimates the reduction in the crop of 1918 in Montana at 25 percent. The disease was first noticed in South Carolina (5) in the form of an unimportant pod spot, but later it became a stem blight which seriously reduced the yield. In Colorado at the time when the blight was first described, losses as high as 50 percent occurred. Dr. Sackett writes that bacterial blight has not been so destructive since.

SYMPTOMS (5, 6, 8,)

Water-soaked spots appear on any or all aerial parts, which are very small at first and finally turn brown to black. The spots may begin near the soil line and extend upward, or they may be scattered. Young flowers and pods may be killed. The spotting of older pods (Fig. 11) may result in internal infection of the seed which may be visible or invisible to the unaided eye.

Fig. 11.—Spots of bacterial blight on pods of the garden pea in various stages of development and coalescence.

CAUSE

The cause of bacterial blight of peas is a micro-organism which was first named *Pseudomonas pisi*, later called *Bacterium pisi*, and is now termed *Phytomonas pisi*. It is a small, swimming, grayish white rod which can enter the plant through stomata and wounds. It can live on pea seeds for 10 months, possibly longer, but it is said not to survive the winter in the soil. In cultures it grows best at approximately 82° F. (28° C.) and it is killed at approximately 122° F. (50° C.). It first enters the field on or in infected seed and is afterward spread by wind, irrigation water, the contact of diseased plants with healthy plants, and probably in other ways.

HOSTS

The work of investigators who have studied bacterial blight of peas shows that the disease can attack cowpeas *(Vigna spp.)*, sweet peas *(Lathyrus odoratus)*, everlasting peas *(Lathyrus latifolius)*, hyacinth bean *(Dolichos lablab)*(8), as well as field and garden peas. It does not affect alfalfa, crimson clover, mammoth clover, sweet clover, and garden beans.

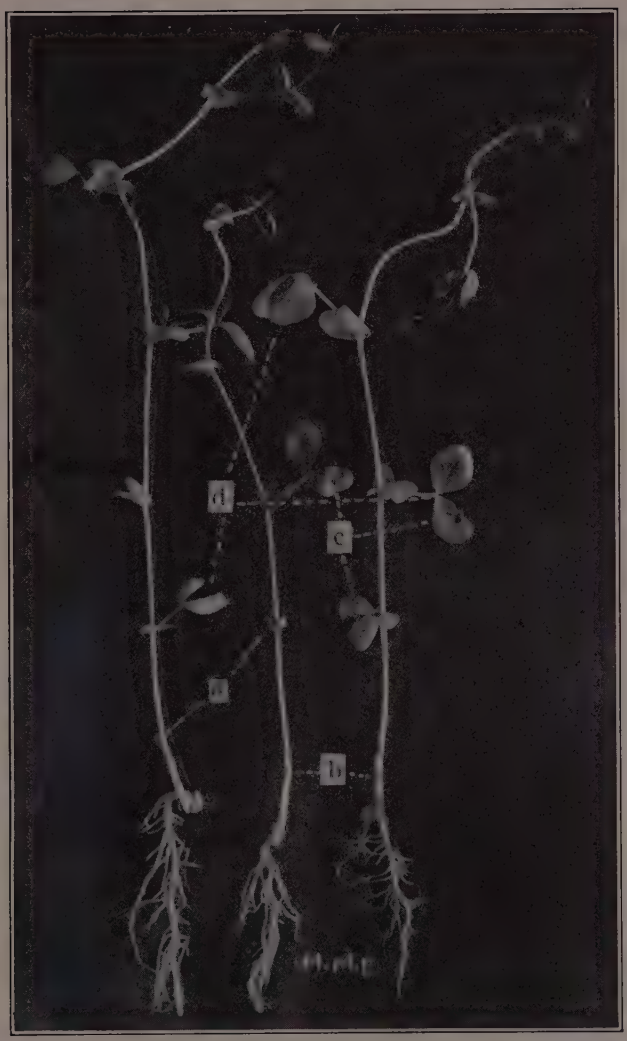

Fig. 12.—Canadian field peas showing early symptoms of bacterial blight caused by *Phytomonas pisi*. The plants were grown from seeds which were immersed in a suspension of the blight bacterium, planted October 30, and the resulting plants were photographed November 24. a. Watersoaked stipules; b. stem lesions; c. leaf lesions; d. yellowing leaves.

BACTERIAL BLIGHT IN ARIZONA

Although bacterial blight of peas has probably been present before in the fields of Arizona, it was first observed in the summer of 1930 and in the same place from which the Fusarium foot rot was obtained. Also the specimens came to us from County Agricultural Agent E. S. Turville. According to Turville the blight affected about 200 acres of peas in the Chino Valley, of which 75 acres were especially bad, since the estimated yield on the latter was not more than 25 percent. The disease was not noticed until the harvesting of green peas was about to commence; then it spread rapidly and soon affected 75 percent of the crop. Later there was a check in the progress of the disease and the new growth was free from the blight.

ORGANISM

The organism which causes bacterial blight of peas, *Phytomonas pisi*, was isolated from the diseased vines and pods sent to us from Chino

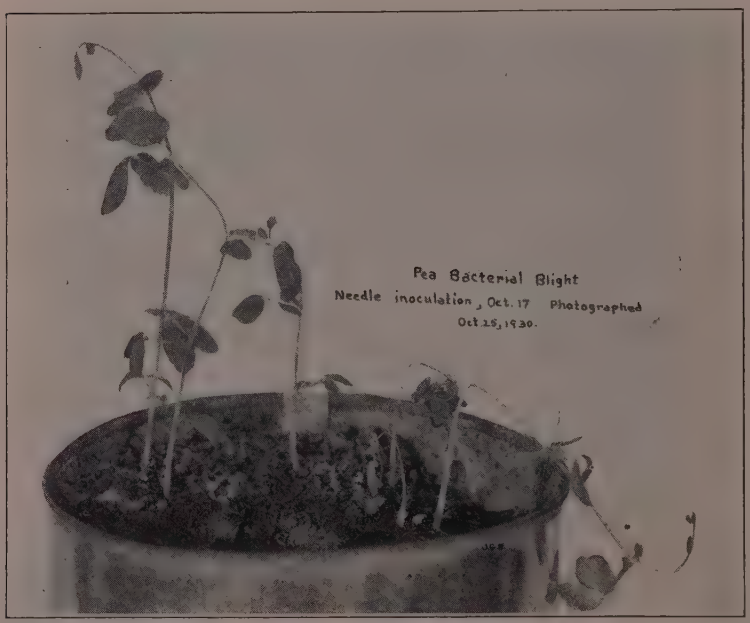

Fig. 13.—Canadian field peas grown from surface-sterilized seed in sterilized soil; controls at left, inoculated plants at right.

Valley, was cultured on numerous media, was inoculated into Canada field peas with resulting symptoms of pea blight (Figs. 12, 13) and was re-isolated. Besides the field peas four varieties of garden peas, namely, Laxtonian, Stratagem, Dwarf Telephone, and Premium Gem, were likewise inoculated with similar results. We also inoculated with the needle young plants of garbanzo *(Cicer arietinum)* which gave local water-soaked spots several millimeters in diameter, but the disease on this plant progressed no further.

Evidence is strong that the infection in Chino Valley came from the seed. No previous crops had been grown on the soil. Furthermore, pea seeds obtained from companies which supply the district with seed were found to be infected and *Phytomonas pisi* was isolated from the infected seed.

Reactions. Since published descriptions of the reactions of *Phytomonas pisi* in general do not include the hydrogen-ion concentrations of the media, we have thought it worth while to record some of the reactions noted in identifying the bacterium. All cultures referred to in the following description were run at 28.5° C.

Medium	24 hours	48 hours	6 days	20 days
Potato—2% dextrose agar streak, pH 6.8.	Fair growth, slightly spreading, lance-shaped, gray, slightly elevated, translucent, moist, smooth, glistening, no discoloration of medium, no odor, no gas.	As before, except light mineral gray, center elevated, margin lobed.	Spoon-shaped, pale gull gray, equally elevated, translucency like very thin porcelain.	
Bacto-nutrient agar, pH 6.8; streak.	Fair growth, nail-shaped, pallid purplish gray, elevated, translucent, moist, glistening, smooth, no discoloration of medium, no odor, no gas.	Luxuriant, lance-shaped, pallid mouse gray; margin crenated and windowed in places.	Like preceding, except pale gull gray.	Pallid neutral gray margin lobed with minute teeth.
Bacto-nutrient agar, pH 7.4; streak.	Like above.	Most luxuriant growth of the three H-ion concentrations tried with nutrient agar; pallid purplish gray.	Margin crenate.	Spreading over most of surface of slant, thin.
Bacto-nutrient agar, pH 7.6; streak.	Scant growth, otherwise like above.	Similar to above, but less growth.	Pale gull gray.	Pallid neutral gray; colony marked with fine transverse lines.
Rice agar, pH 6.8; streak.	Slight growth.	Slightly spreading, scant, pale gray, elevated, translucent, moist, glistening, no discoloration of medium, no odor, no gas.	Club-shaped, scant, gray, elevated, translucent, moist, smooth, glistening, no discoloration of medium, no odor, no gas.	Club-shaped, very thin, surface etched with fine transverse lines; same gray color as agar.
Corn-meal agar, pH 6.8; streak.	Scant growth.	Same as rice agar.	Similar to rice agar, but very pale yellow tinge.	Like rice agar, but faint trace of yellow.

Medium	24 hours	48 hours	6 days	20 days
Potato plug.	Spreading slightly, needle-shaped; scant, pale yellow growth, slightly elevated, moist, glistening, no discoloration, no odor, no gas.	Fair growth, spreading, elevated, maize yellow, almost opaque, moist, smooth, glistening, no discoloration, no odor, no gas.	Fair growth, spreading, club-shaped, chamois color, elevated, moist, smooth, glistening, no discoloration, no odor, no gas.	Tawny olive to snuff-brown.
Nutrient gelatin stab, pH 7.4; room temperature.	(27 days) Half liquefied, no gas.			
Dextrose-gelatin, pH 7.4.;	(21 days) Complete liquefaction in 21 days; a few gas bubbles; dense gray, viscid precipitate below liquid.			
Nutrient broth.	No growth.		Slight growth, small quantity of dark brown precipitate.	
Litmus milk.	No change.	No change.	Litmus reduced; whey turbid; liquefaction of coagulum; no gas; pale straw color; ring robin's egg blue; no odor.	One inch of clear whey above bottom of tube; coagulum almost completely peptonized; cream-buff precipitate; ring pale glaucus-blue.
Uschinsky	No growth.	No growth.	No growth.	No growth.
Dextrose, 1%	No growth.	Cloudy in open arm.	Cloudy only in open arm; no gas; heavy gray precipitate.	
Lactose, 1%	No growth.	No growth.	No growth.	
Maltose, 1%	No growth.	Cloudy in open arm, but less so than dextrose.	Good growth in open arm; none in closed; no gas.	
Sucrose, 1%	No growth.	Cloudy in open arm only.	Cloudy only in open arm; no gas.	

SUMMARY

1. This paper describes a destructive foot rot which attacked peas planted in virgin alkaline soil in central Arizona.

2. From the infected pea plants a Fusarium was isolated, grown in pure cultures from a monosporidial source, inoculated into healthy pea plants in which it caused the foot rot, and re-isolated.

3. The reactions of pure cultures of the Fusarium on various media are given; also a description of the mycelium and spores, and the identification of the fungus.

4. A few observations are recorded on the cultural reactions of bacterial blight *(Phytomonas [Pseudomonas] pisi* Sackett), a second pea-disease previously unreported from Arizona.

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ACKNOWLEDGMENTS

The cooperation of County Agricultural Agent E. S. Turville of Yavapai County, who brought to our attention the presence of foot rot and bacterial blight of peas in Arizona and supplied field specimens for our study, is appreciated. We are also indebted to Dr. R. B. Streets, Associate Pathologist in this Department, for the photographs which bear his initials.

FUSARIUM FOOT ROT OF PEAS

Studies of Cultures of Monosporidial Origin

All drawings were made with the aid of a camera lucida and, excepting Figures 1, 23, and 24, with Spencer apochromatic ocular ×15 and 2-millimeter oil-immersion objective N.A. 1.40.

PLATE I

Magnifications: Figure 1 reduced to ×515; others to ×1072.

Fig. 1.—Hypha from 23-day culture of autoclaved rice, to show branching.

Fig. 2.—Anastomoses of hyphal cells, from 27-day culture on autoclaved potato plug.

Fig. 3.—Swollen cells, 14-day culture on potato-2 percent dextrose agar.

Fig. 4.—Parts of hyphae showing (a) thick walled, intercalary resting cells and (b) swollen intercalary cells and young branches; from 34-day culture on autoclaved potato plug.

Fig. 5.—Branching of swollen hyphal cell, from 6-weeks culture on autoclaved potato plug.

Fig. 6.—Cross-section of swollen cell stained with iron-alum haematoxylon; from culture on autoclaved potato plug.

Fig. 7.—Branching of conidiophore; from culture on autoclaved potato plug.

Fig. 8.—Anastomosis of macroconidia, from 20-day culture on potato-2 percent dextrose agar.

Fig. 9.—Anastomoses of macroconidia *en masse*, from 20-day culture on potato-2 percent dextrose agar.

Fig. 10.—Anastomosis of macroconidium with hypha, from 9-day culture on potato-2 percent dextrose agar.

Fig. 11.—Macroconidium in long section, stained with iron-alum haemotoxylon; from young culture on autoclaved potato plug.

Fig. 12.—Resting cell within macroconidium, from culture on autoclaved potato plug.

Fig. 13.—Germination of macroconidium; effect of food reserve on vigor of germ tube.

Fig. 14.—Branching of swollen cell from fragment of hypha, from 81-day culture on autoclaved potato plug.

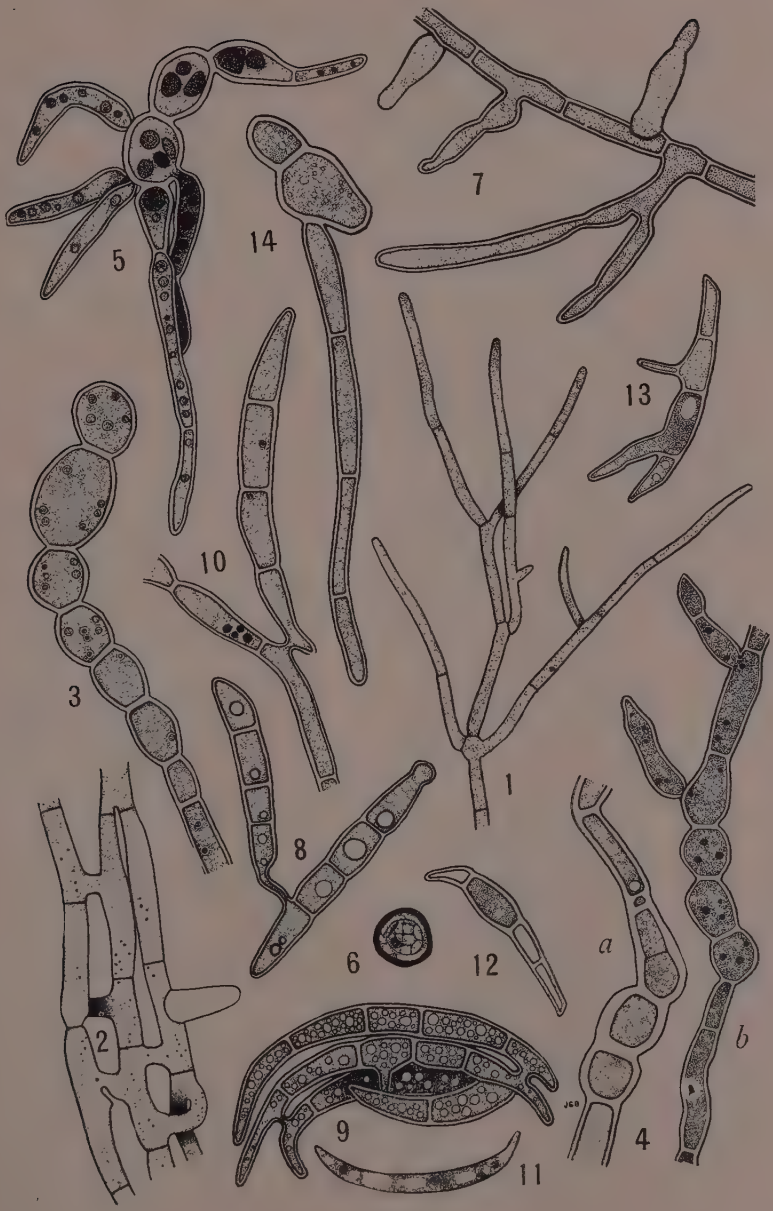

PLATE I

PLATE II

Forms of Conidia

Magnification reduced to ×1210

Fig. 15.—Conidia from culture on autoclaved potato plug: (a) 6 weeks old; others 1 week old.

Fig. 16.—One-septate conidia from culture on autoclaved potato plug: (b) 1 week old; (c) 81 days old; others 6 weeks old.

Fig. 17.—One-septate conidia showing range of forms from 3-weeks culture on autoclaved potato plug.

Fig. 18.—Two-septate conidia from culture on autoclaved potato plug: (d) 1 week old; (e) 6 weeks old.

Fig. 19.—Three-septate conidia: (f) from 1-week culture on autoclaved potato plug; (g) from 9-day culture on potato-2 percent dextrose agar, pH 7; others from 81-day culture on autoclaved potato plug.

Fig. 20.—Four-septate conidia: (h) from 6-weeks culture on autoclaved potato plug; (i) from 81-day culture on potato-2 percent dextrose agar, pH 7.

Fig. 21.—Five-septate conidia: (j) from 6-weeks culture on autoclaved potato plug; (k) from culture on autoclaved potato plug; (l) from 81-day culture on potato-2 percent dextrose agar, pH 7.

Fig. 22.—Six-septate conidium with resting cells.

PLATE II

PLATE III

Conidiophores, Spores, and Swollen Cells

Magnification reduced to ×543 in Figures 23 and 24; to ×1063 in Figures 33 and 34; to ×1066 in the other drawings.

Figs. 23, 24.—Conidiophores from 8-day culture on proteose-peptone agar, pH 7.

Figs. 25, 26.—Conidiophores to show details of branching; from 9-day culture on proteose-peptone agar, pH 7.

Fig. 27.—Non-septate conidia from 9-day culture on proteose-peptone agar, pH 7.

Fig. 28.—One-septate conidia from 9-day culture on proteose-peptone agar, pH 7.

Fig. 29.—Two-septate conidia from 9-day culture on proteose-peptone agar, pH 7.

Fig. 30.—Three-septate conidia: (a) from 5-day culture; (b) from 9-day culture; others from 11-day culture on proteose-peptone agar, pH7.

Fig. 31.—Four-septate conidia: (a) from 9-day culture; (b) from 11-day culture; proteose-peptone agar, pH 7.

Fig. 32.—Five-septate conidium from 9-day culture, proteose-peptone, pH7.

Fig. 33.—Terminal chlamydospores from 404-day culture, nutrient agar, pH 7.

Fig. 34.—Group of chlamydospores from 405-day culture on nutrient agar, pH 7.

Fig. 35.—Swollen cells: (a) much vacuolated; (b, c) in later stages of degeneration; from culture on autoclaved potato plug; sectioned and stained with iron-alum haematoxylon.

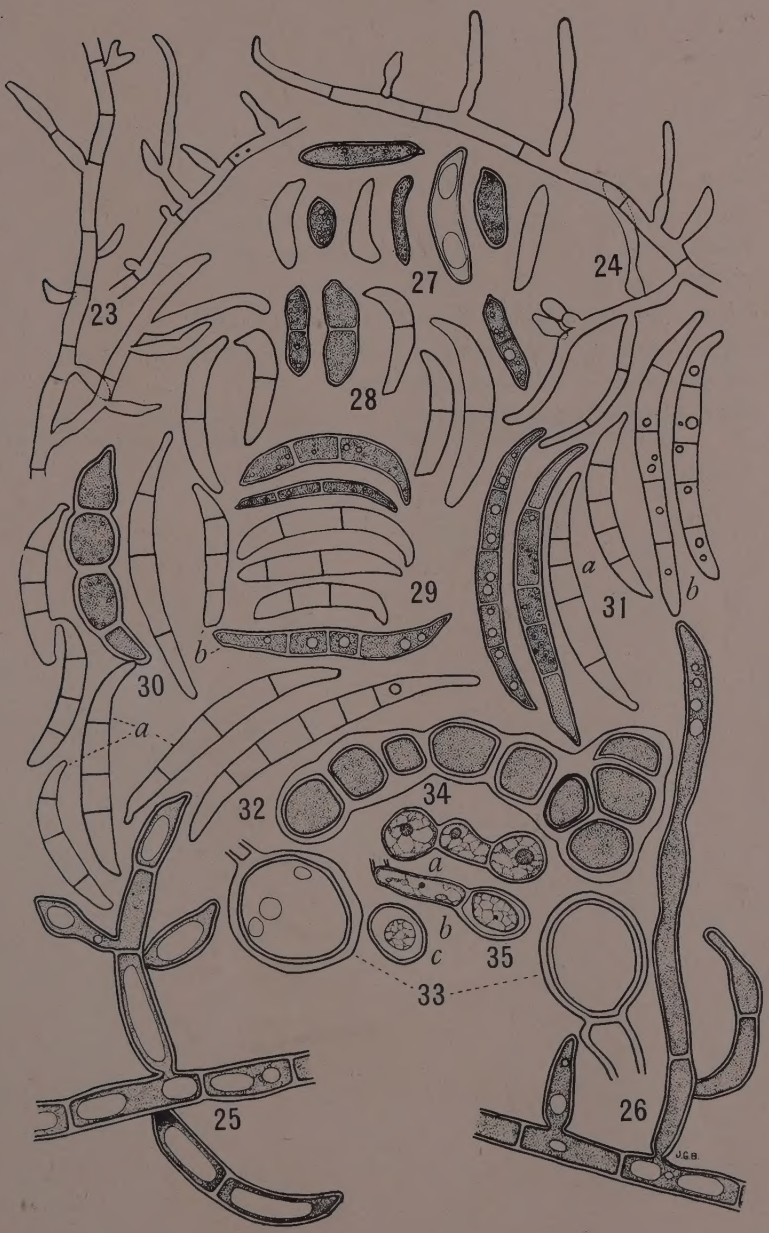

PLATE III

PLATE IV

All photographs X3

Fig. 36.—Three-day culture on proteose-peptone agar, pH 7.

Fig. 37.—Seven-day culture on proteose-peptone agar, pH 7.

Fig. 38.—Seven-day culture on proteose-peptone agar, pH 5.2.

Fig. 39.—Three-day culture on nutrient agar, pH 7.0.

Fig. 40.—Three-day culture on potato-2% dextrose agar, pH 7.

Fig. 41.—Three-day culture on Czapek's solution agar, pH 5.6.

Fig. 42.—Culture on autoclaved walnut stem.

Fig. 43.—Vertical section through culture on autoclaved potato plug. a, pionnotes; b, loose network of stromatal layer; c, denser layer of stroma; d, medium.

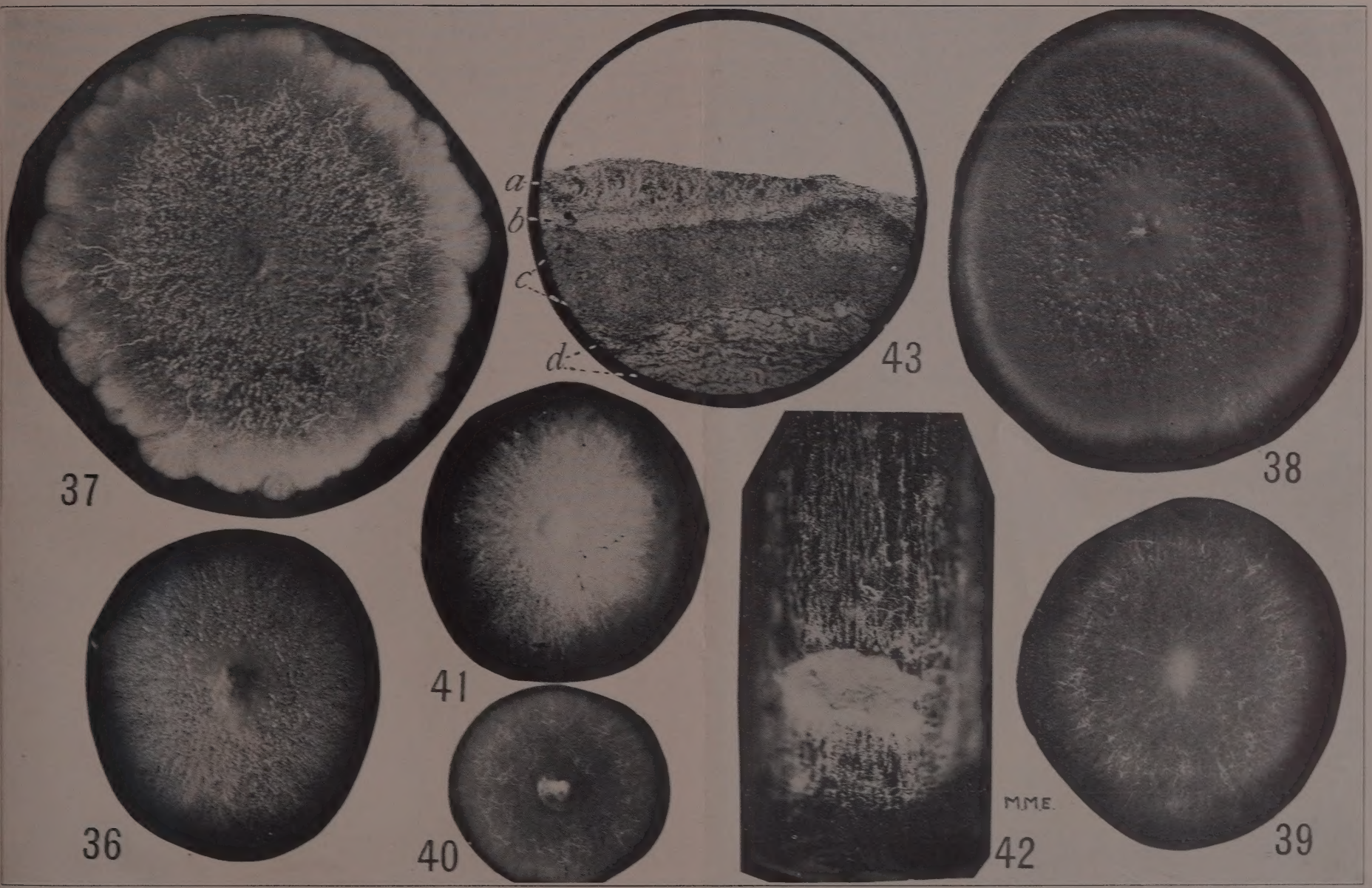

PLATE IV